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Larval development and feeding ecology of Hermissenda crassicornis (Eschscholtz) and Aeolidia papillosa (Linnaeus) : a thesis ...

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LARVAL DEVELOPMENT AND FEEDING ECOLOGY OF *HERMISSENDA CRASSICORNIS* (ESCHSCHOLTZ) AND *AEOLIDIA PAPILLOSA* (LINNAEUS)

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by

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INTRODUCTION

Thompson (1967) defines three developmental types for the Opisthobranchia with representative nudibranchs in each category. The types are: 1.) planktotrophic larvae which are obligatory plankton feeders prior to progressive metamorphosis, 2.) lecithotrophic larvae which may feed on the plankton, but do not need to do so in order to metamorphose, 3.) direct development which results in hatching of a post veliger, benthic juvenile.

Tardy (1970) feels that Thompson's Type 3 development is artificial. Thus, he incorporates direct development into the lecithotrophic developmental type. Tardy then proposes a classification of metamorphic types based on larval shell type (Thompson, 1961) and on larval feeding behavior (ie. lecithotrophic vs. planktotrophic larvae). I agree with Thompson's (1967) distinction between direct and lecithotrophic developmental types in the sense that they represent ecologically diverse ontogenies.

The major explicit assumption in the above definition of planktotrophic larvae is that feeding is a necessary prerequisite to progressive metamorphosis. However, both Thompson (1967) and Tardy (1970) appropriately note that metamorphic observations of planktotrophic larvae are fragmentary and circumstantial. Thus, the definition of planktotrophic larvae rests its creditability on larval morphology

at hatching, and the implicit assumption that energy is required (ie. feeding) to develop the organs necessary to accommodate the functional transition to the adult mode of life.

Thus, Thompson's (1958, 1962, 1967) and Tardy's (1970) definitive works with respect to the description and classification of developmental and metamorphic types are implicitly bound to the definition of larval feeding requirements. Unfortunately all of these cited works utilized lecithotrophic or directly developing larvae as their primary source of observations. It remains, therefore, to establish the relationship between planktotrophic larval morphology and larval feeding requirements within the context of evolutionary and ecologically distinct aspects of life history variation.

Thompson (1967, p. 16) concludes:

> "The majority of species, from all taxonomic subgroups of the Opisthobranchia, possess planktotrophic veliger larvae. A smaller number of species from various subgroups, give rise to lecithotrophic larvae; the pressures which have led them to adopt this type of life history are unknown: no obvious common ecological or taxonomic features unite these species. Direct development is exhibited by species which are in the main of small adult size. . . and species having a boreo-arctic distribution. Doridopsis limbata and Eolidina alderi are, however, examples whose significance is at present unknown."

It is precisely the lack of common taxonomic features uniting the three ecologically diverse developmental types which is significant in considering the evolution of Nudibranch larvae. Tinkle (1969, p. 501) states that,

> "Most aspects of the life histories of individual organisms are explicable in terms of natural selection, a fact often neglected in life history studies, with the result that such studies rarely contain the type of information necessary to test evolutionary hypotheses concerning life history phenomena."

For instance, there is considerable developmental variability within taxonomically conservative groups; the genus Aeolidiella contains species representative of all three developmental types (Thompson, 1967). Secondly, there is evidence pointing to the occurrence of divergent developmental types within species: Dendronotus frondosus (Ascanius) (Thompson, 1967; Williams, In Press).

The present study describes the early embryology, larval development, veliger morphology and feeding behavior of Hermissenda crassicornis and Aeolidia papillosa. Veligers of the facelinid, Hermissenda crassicornis and the aeolid, Aeolidia papillosa have striated, Type 1 (coiled) larval shells and fall within Thompson's (1967) definition of planktotrophic larvae. Veligers of both species remain planktonic for two to five days after hatching. They subsequently become epibenthic swimmers and discard their larval shells. There is considerable variation in the amount of yolk reserves in the gut and diverticulae of recently hatched veligers. An individual egg mass yields larvae with and without yolk reserves.

The results show that shell length frequency of hatching larvae is distributed bimodally. There is a larval dimorphism

based on shell length at hatching, the presence of yolk reserves and feeding ability. Feeding larvae are found to differ with respect to diet; the difference being associated with shell length of the larvae in relation to food particle size. The results are discussed in a comparative review of larval development in the Eolidoidea. Secondly, the relative dependancy of nudibranch larvae on feeding ability is discussed with respect to the morphological and developmental categorization of opisthobranch metamorphic responses. The functional and ecological considerations of feeding in gastropod and selected invertebrate larvae are discussed with respect to the evolution of larval strategy and life cycle phenomena.

MATERIALS AND METHODS

Adult *H. crassicornis* were collected from *Zostera marina* L. beds located on Lawson's Flat, Tomales Bay, California. Adult *A. papillosa* were collected from the boat floats at Mason's Marina and from the North Jetty in Bodega Harbor, California. Adults of both species were isolated in plastic aquaria, and supplied with running seawater in the laboratory.

Adults of both species readily spawned in the laboratory from August, 1970 through July, 1971. Development was followed for 50 *H. crassicornis* and 25 *A. papillosa* egg masses spawned and cultured in the laboratory. The extreme abundance of *H. crassicornis* and its spawn on Lawson's Flat allowed additional observations on approximately 120 egg masses collected in the field.

Egg masses deposited on the sides of aquaria were removed and cultured in 500 ml flasks at 14.0°C. The egg masses and hatched veligers were washed daily on a 45 μ mesh screen, and returned to clean culture flasks containing filtered seawater. Hatched larvae were fed mixed suspensions from algal cultures of *Monochrysis lutheri* Droop, *Isochrysis galbana* Parke, *Dunaliella tertiolecta* Butcher, *Chlorella* 580, and *Phaeodactylum tricornutum* Bohlin. Algae were cultured in a growth chamber at 17°C. Algal cultures were grown in Guillard and Ryther's (1962) f_2 medium.

Random samples (N-100) of larval shell length were taken to delimit the shell length frequency distribution of recently hatched larvae. F-Max and Runs tests indicate that within species, variances are homoscedastic, and that shell length observations are random and independant (See Sokal and Rohlf, 1969).

In the case of H. crassicornis larvae, three feeding experiments were conducted. The first was a qualitative appraisal to determine which of the above algal species the veliger larvae would accept as food.

The second dealt with the relative clearance rates of algae retained in the left digestive diverticulum after feeding. In this instance, recently hatched larvae were placed in a combined suspension of Monochrysis, Isochrysis, and Dunaliella. After 48 hours the larvae were removed to clean, filtered seawater. 700 larvae were sampled (N=100) over a 53 hour period, and examined for the presence of algal cells or pigmentation in the left digestive diverticulum.

Finally, a feeding experiment was designed to test the feeding ability of yolked and non-yolked veligers on two species of algae. Two flasks containing 500 mls of sterile, filtered seawater, and f_2 media were used in the experiment. Salinity in each flask, after addition of the media, was 33.0°/oo. One flask contained a suspension of Dunaliella, while the second flask contained a suspension of Chlorella 580. Both flasks contained approximately 1,000 recently hatched H. crassicornis

veligers. The flasks were placed in a refrigerator set at 14.0°C. ± 0.5°C. Constant illumination was provided during the experiment with a 40 watt daylight-type flourescent lamp. The flasks were left in the refrigerator for 72 hours.

At the initiation of the experiment, 100 additional larvae were sampled from the egg mass and examined for the presence of yolk reserves in the left diverticulum. Shell length measurements were then recorded for 20 yolked and 20 non-yolked larvae.

At the termination of the experiment, the flasks were removed from the refrigerator and the larvae gently washed on the 45 μ mesh screen. From each of the algal test groups, 100 larvae were examined for the presence of algae in the left diverticulum. Secondly, shell lengths were recorded for 20 larvae with food in the diverticulum and for 20 larvae without food in the diverticulum, for each algal test troup. Finally, an analysis of variance utilizing partioning of mean square and standard 'F' tests was performed on the shell length data.

In the case of A. papillosa veligers, a similar series of experiments were performed, but with the following exceptions. Three species of algae were used in the experiment testing the relative feeding ability of yolked and non-yolked larvae: Chlorella 580, Dunaliella, and Phaeodactylum. At the initiation of the experiment, 50 yolked and 50 non-yolked larvae were sampled for shell length. In addition to the three monospecific test flasks, a fourth test flask containing a

mixed suspension of Chlorella 580 and Phaeodactylum was employed. At the termination of the experiment shell lengths were recorded for 30 feeding and 30 non-feeding larvae from each test flask.

The objectives underlying the feeding experiments and subsequent analyses of variance were to demonstrate possible larval dimorphism delineated by the presence of yolk reserves, larval shell length, and feeding ability. Secondly, it was hypothesized that veliger feeding ability is influenced by particle size selectivity. Statistical analysis followed the procedure outlined by Sokal and Rohlf (1969).

Observations were made on living material with a compound microscope utilizing phase contrast and bright field optics. Still photomicrographs were taken with a 35 mm camera, and motion pictures with a 16 mm camera. The 16 mm motion pictures were used to analyze rotational movement of H. crassicornis veligers.

RESULTS

Egg Masses:

Both Hermissenda crassicornis and Aeolidia papillosa have white, spirally coiled type B egg masses with secondarily twisted egg strings (Hurst, 1967; Figure 1). The H. crassicornis and A. papillosa egg strings consist of individual egg capsules. The H. crassicornis egg capsules are elliptical and non-wrinkled. A. papillosa egg capsules are irregularly shaped and have a wrinkled appearance.

Occasionally, the egg capsules of both species do not separate during spawning and are, therefore, joined end to end by the capsule membrane. In H. crassicornis the portion of the capsule membrane connecting egg capsules is a simple constriction. In A. papillosa, however, the capsule membrane connecting egg capsules is twisted, as well as, constricted (Figure 2).

Hurst (1967) observes that the numbers of ova per egg capsule for A. papillosa is rather variable, but fairly constant within one egg mass. An analysis of variance for the number of ova per egg capsule sampled from 17 egg masses spawned by ten H. crassicornis corroborates Hurst's generalizations for A. papillosa. Thus, in H. crassicornis there is a significant added variance component among egg masses for the number of ova per egg capsule ($p<0.01$). The coefficient of intraclass correlation is 0.535. The source of intraclass

variation is apparently twofold. Successive spawning by the same individual often result in fewer ova per egg capsule. In some instances, the apparent depletion of ova results in the formation of empty egg capsules at the terminal end of the egg string. Secondly, larger individuals tend to produce more ova per egg capsule than do smaller individuals.

Gametes:

The ova of H. crassicornis and A. papillosa are small. The mean diameter of H. crassicornis ova is 64.7 $\mu \pm$ 1.6 while the mean diameter of A. papillosa ova is 73.7 $\mu \pm$ 2.2 (Figure 3). Analysis of variance indicates a significant ($p \ll 0.001$) added variance component between zygote diameters for the two species. The coefficient of intraclass variation is 0.95. Thus, the greatest proportion of the variance is between species. Length of the spermatozoa in both species is approximately 150 μ.

Development to Hatching:

The major developmental features and developmental rates to hatching for H. crassicornis and A. papillosa are summarized in Table I.

The separation of the polar bodies from the ova occurs at the animal pole in both H. crassicornis and A. papillosa. Completion of the maturation divisions results in the formation of two polar bodies for both species. A. papillosa, however, is characterized by the division of the first polar

body shortly after the first cleavage (Figure 4). The polar bodies remain intact through early veliger and often through hatching.

Cleavage is similar in H. crassicornis and A. papillosa. The first cleavage is holoblastic and equal, with the cleavage plane laying in the polar axis. The second cleavage is asynchronous, holoblastic and equal, with the second cleavage plane laying in the polar axis at right angles to the first cleavage plane. Second cleavage is characterized by the holoblastic division of one of the blastomeres to form a transitory three cell stage (Figure 5). The second blastomere divides approximately ten minutes after the first. When viewed from the animal pole, the division of the second blastomere is characterized by the counter-clockwise displacement of the daughter blastomere into a slightly lower focal plane. Thus, the second cleavage appears to be laeotropic. The resultant four cell stage is distinguished by having the animal and vegetal polar furrows oriented at right angles to one another. The third cleavage is dexiotropic, holoblastic and unequal. The fourth and ensuing cleavage divisions are typical of the molluscan pattern of alternating spiral cleavage.

Cleavage in both species results in the formation of a stereoblastula (Figure 6). Gastrulation occurs by epiboly and is characterized by the formation of a blastoporal sagittal cleft. During late gastrula, the sagittal cleft closes in a zipper-like fashion beginning near the animal pole

and terminating with the closure of the definitive blastopore at the vegetal pole. Gastrulation results in a stereogastrula (Figure 7). Other workers concerned with early embryology in the Opisthobranchia have described a coelogastrula (Rasmussen, 1951; Rao and Alagarswami, 1960). These illustrations, however, depict a sagittal cleft as described and illustrated by Raven (1958).

Following gastrulation, the trochophore stage is recognized by the development of an anterior ciliary crown, the prototroch. In each species, the appearance of the prototroch is rapidly followed by the formation of two anal cells posteriorly, and the shell gland invagination left posterolaterally (Figure 8). Subsequently, the prototroch differentiates into two rudimentary velar lobes, the shell gland evaginates, and the metapodium forms. As the shell gland spreads to circumscribe the cephalopedal complex, the anal cells are displaced anterolaterally to the right side. The developing embryos are, at this point, early veliger larvae.

Next the shell gland secretes the definitive early veliger shell and, then, differentiates into the mantle fold and the perivisceral membrane.

The transition to the fully developed pre-hatching veliger is characterized by continued shell growth, the stomodeal invagination, differentiation of the alimentary and larval excretory organs, formation of retractor and mantle

muscles, the deposition of an operculum and the growth of "tactile" pedal cilia (Figure 9).

The larval shell in both H. crassicornis and A. papillosa veligers is characterized by oblique striations on the shell whorl. These striations are present at the initial formation of the larval shell. Thus, in the early veliger the striations extend from the ventral surface of the whorl posterolaterally to the right dorsolateral surface of the shell. However, in the completely differentiated veliger, the striations extend from the junction of the aperature with the whorl left posterolaterally to the ventral surface of the whorl. This demonstrates the differential shell growth rate along the mantle fold-shell aperature junction and accounts for the hyperstrophic shell. The left anterolateral margin of the shell grows more rapidly than the right margin. The shell, then, appears to be sinistral while the internal organization of the larvae is dextral.

Eclosion:

Hatching of the mature veligers occurs similarly in H. crassicornis and A. papillosa. As the veligers near hatching, they become increasingly active. The capsule membrane becomes thin and flacid. In the case of A. papillosa, the egg capsule loses its wrinkled appearance. The larvae vigorously beat their velar cilia against the capsule wall. The effective stroke of the velar cilia is forwards over the velum. The direction of movement within the capsule is shell first or

posterior. Hatching occurs by rupture of the capsule wall along the larval shell-capsule membrane interface. The veliger exits the capsule by backing through the rupture.

Free Swimming Veligers:

Free swimming H. crassicornis and A. papillosa larvae exhibit almost identical gross morphological characteristics at hatching. The larval shell for both species is the coiled, non-inflated variety as defined by Thompson (1961). The shells demonstrate hyperstrophic assymetry. As mentioned above, the shells are characterized by six to seven oblique striations on the whorl. The means and standard deviations for shell length, height, and width are summarized in Table II. Shell length:width:height ratios are 1.276:1:0.972 for H. crassicornis and 1.355:1:0.977 for A. papillosa.

The cephalopedal regions are characterized by moderate sized velar lobes, prominent sub-velar ridges, and bristle-like lateral and apical 'tactile' foot cilia.

Internal organization is dextral with the anal aperture opening into the right side of the mantle cavity immediately behind the larval kidney. The intestine of both species is distinguished by having a warty appearance. The larvae concur with Thompson's (1967) definition of planktotrophic veligers. They lack eyes, radula, propodial rudiment, and adult kidney vesicle. They possess, however, a pair of nephrocysts in addition to the larval kidney.

The differences between the two species are quantitative rather than qualitative. Shell length frequency distributions, for both species, are bimodal (Figure 10 and Figure 11).

The bimodal shell length frequency distributions may be explained in light of veliger dimorphism with respect to the amount of yolk reserves present at hatching. Thus, 40%-60% of recently hatched H. crassicornis and 80%-90% of recently hatched A. papillosa larvae have yolk reserves in the gut and/or left digestive diverticulum.

Mean shell lengths are 103.8 μ $\pm$ 8.4 and 116.7 μ $\pm$ 11.0 for the larvae of H. crassicornis and A. papillosa respectively. The bimodal distribution peaks for H. crassicornis larvae are at 93.8 μ and 107.3 μ. The distribution peaks for A. papillosa larvae are at 107.3 μ and 120.8 μ. Mean shell lengths and 95% confidence intervals based on the 't' distribution are shown in Figure 19.

Cephalopedal Ciliation:

The mechanical treatment of food is similar for both H. crassicornis and A. papillosa veligers. The discussion of cephalopedal and internal ciliation is therefore applicable to both species.

Cephalopedal cilia may be categorized as follows: 1.) velar cilia, 2.) post oral cilia located on the sub-velar ridge, 3.) oral cilia, 4.) pedal cilia, and 5.) lateral and apical "tactile" cilia located at the junction of the foot and operculum (Figure 9).

During active unidirectional swimming, the velar cilia beat in a regular, diaplectic, metachronal fashion. If viewed from the ventral side of the veliger, the metachronal waves of both velar lobes move in a clockwise direction (Figure 12). The effective stroke is backwards over the velum. The velar cilia, therefore, show laeoplectic metachronism (Knight-Jones, 1954). Thus, water, as well as, suspended particulate matter, is displaced laterally and posteriorly over the velum. The direction of movement, is velum first with shell and visceral mass trailing.

The velar cilia are active in effecting motion other than the unidirectional forward movement described above. Hurst (1967) observed right handed circular movement and backwards axial spinning for H. crassicornis veligers. During circular movement, she noted a partial withdrawal of the right velar lobe, as well as, the extension of the velar cilia on the right lobe. Observations in the present study agree with Hurst's description of the right velar lobe and cilia. However, the rotation does not appear to be regularly circular. Ciné photomicrographs reveal three separate directional components in circling movement: 1.) right antero-lateral, 2.) right handed axial spinning, and 3.) posteriorly directed motion (Figure 13).

The effective stroke of the post-oral cilia, on both velar lobes, is towards the mouth. Suspended particles transported over the velum by the velar cilia are caught

by the post-oral cilia and passed to the mouth. The oral cilia either manipulate the particulate matter into the esophagus, or pass it to the anterior base of the foot.

The effective stroke of the pedal cilia is towards the apex of the foot. Thus, particles rejected by the oral cilia are conducted away from the larvae via the long axis of the foot. The pedal cilia also function in the rejection of particles displaced over the velum, but not accepted by the post-oral cilia (Figure 14). On the right posterolateral margin of the foot, a strong rejection current passes faeces and whole undigested algal cells away from the anus to the apex of the foot.

No apparent functional utilization of the bristle-like "tactile" cilia was observed.

Internal Ciliation:

Algal cells accepted by the oral cilia are transported through the esophagus to the ventral stomach region (Figure 15). Food particles are actively rotated counter-clockwise (dorsal view) against denticulate hyaline rods situated in the posteroventral stomach wall. Food material is then passed into the left diverticulum. Particle rotation within the diverticulum is clockwise. Quite often, large particles are passed out of the diverticulum, recirculated in the ventral stomach, and returned to the diverticulum. It must be emphasized, that particle transport between the ventral gut and the diverticulum is achieved by ciliary manipulation.

Particles not broken up in the ventral stomach, nor accepted by the left diverticulum, are moved against the anterior wall of the stomach and rapidly passed along the intestinal groove to the intestine. Smaller, broken up algal cells are slowly rotated in a counter-clockwise spiral through the mid gut and dorsal gut. Faecal matter is loosely compacted in the dorsal gut and passed into the intestine.

Metachronal waves in the intestine move from the anus toward the gut. The effective stroke of the cilia is towards the anus. Intestinal cilia behave in an antiplectic fashion (Knight-Jones, 1954). Thus, faeces and whole undigested algal cells are moved along the intestine, excreted through the anus, and conducted away from the larva along the pedal rejection current.

Feeding in H. crassicornis Larvae:

H. crassicornis veligers readily accept as food the algal species Monochrysis, Isochrysis, Chlorella 580, and Dunaliella. Mean diameters (length for Dunaliella) and standard deviations for these species are shown in Table III.

Provided the gut and left diverticulum are yolk free, just hatched larvae feed voraciously when placed in a suspension of any of the above algal species. The gut, the left diverticulum, and the esophagus are filled with algal cells within ten minutes after the addition of a heavy algal suspension. In cases of over feeding, veligers cease to feed temporarily. Overfed larvae defecate undigested

Algal cells until the esophagus is cleared and algal cells are found only in the gut and diverticulum. Thereafter, feeding occurs sporadically; most algal cells are either passed over the velum, or are rejected by the oral cilia. In cases where the larvae are not initially overfed, veligers feed actively until both the gut and the diverticulum contain algal cells. Feeding then occurs sporadically, as described above.

The sample data for gut clearance rate is plotted in Figure 16. Approximately 80% of the larvae which had fed retained algal cells in the left diverticulum for 53 hours after removal from the suspension. Thus, the initial rate of particle ingestion appears to accomodate the physical capacity of the larval gut to retain algal particles, while the subsequent, occasional, feeding most likely accomodates the rates of digestion and excretion.

The relative numbers of yolky and of feeding larvae are shown in Figure 17. There are relatively fewer larvae feeding on the algal suspensions than there are non-yolky larvae at hatching. Secondly, the relative numbers of non-feeding larvae to feeding larvae is the same for both the Chlorella 580 and the Dunaliella tertiolecta suspensions.

Mean shell lengths and 95% confidence intervals for the experimental larvae are presented in Figure 18a. The analysis of variance and a priori tests for the experimental larvae are summarized in Table IV.

Thus, there is a highly significant added variance component in shell length between yolky and non-yolky larvae, as well as, between feeding and non-feeding larvae (Figure 19 and Figure 20). Secondly, there is no significant difference in shell length between yolky and non-feeding larvae, nor between non-yolky and feeding larvae. These results suggest that feeding ability is curtailed by the presence of yolk reserves in the left diverticulum at hatching.

However, if the null hypothesis is accepted, that there is no difference in shell length between non-yolky and feeding larvae, how can one account for the difference in relative numbers of non-yolky larvae at hatching as opposed to the relative numbers of feeding larvae at 72 hours after hatching (Figure 17)? The difference may be explained in light of the significant added variance component in shell length between larvae feeding on Chlorella 580 and larvae feeding on Dunaliella. The alternative hypothesis, in this case, is that larger larvae select larger food particles (viz. Dunaliella), while smaller larvae select smaller food particles (viz. Chlorella). Thus, in each test flask not all of the larvae capable of feeding were successful because of particle size selectivity. This argument may be corroborated by the sample shell length distributions. Larvae feeding on Chlorella are skewed left with the mean at 104.4 μ. Larvae feeding on Dunaliella are skewed right with the mean at 108.4 μ (Figure 20).

These observations, however, do not obviate the possibility that particle selectivity may be a function of veliger

size rather than particle size. That is, apparent particle size selectivity may represent a mechanical limitation of the feeding/ingestion apparatus, rather than a size preference geared to the largest particle size most efficiently consumed. However, these two possible explanations for selectivity are reciprocal in nature; thus yielding the same net effect.

Feeding in Aeolidia papillosa Larvae:

The relative percentages of non-yolky and feeding larvae are shown in Figure 17. Unlike H. crassicornis larvae, there is little discrepancy between the relative numbers of non-yolky and feeding larvae. Finally, the proportion of non-yolky to yolky and of non-feeding to feeding larvae is significantly less than for H. crassicornis larvae. Mean shell lengths and 95% confidence intervals for A. papillosa experimental larvae are shown in Figure 18b. The analysis of variance for larval shell lengths is summarized in Table V.

In the case of A. papillosa larvae, there is a highly significant added variance component ($p<<0.001$) in veliger shell length between yolky and non-yolky larvae, as well as, between feeding and non-feeding larvae. There is no significant difference in shell length between yolky and non-feeding larvae (Figure 19 and Figure 21). However, there is a significant added variance component ($0.001<p<0.005$) in larval shell length between non-yolky and feeding larvae. These results suggest the following alternative hypotheses: 1.) the mean shell length of non-yolky larvae is greater than yolky

larvae; 2.) the mean shell length of feeding larvae is greater than non-feeding larvae; 3.) the presence of yolk reserves in the gut and/or left diverticulum does not necessarily pre-empt feeding ability.

Figure 21 illustrates the sample shell length frequency distributions of feeding and non-feeding larvae respectively. The null hypothesis is confirmed in two way comparisons of the means between larvae feeding on Chlorella and larvae feeding on Dunaliella, as well as, between larvae feeding on Dunaliella and larvae feeding on Phaeodactylum. There is a significant ($0.001<p<0.005$) added variance component in larval shell length between larvae feeding on Chlorella and larvae feeding on Phaeodactylum. The analysis of variance for feeding larvae and the progressively increasing mean shell length of larvae fed successively larger algal particles (Figure 18) suggest two hypotheses with respect to feeding behavior: 1.) Smaller larvae are physically incapable of feeding on larger algal particles (viz. Phaeodactylum); or 2.) Larger larvae select larger algal particles.

Feeding larvae from the unialgal suspensions have skewed left sample shell length frequency distributions. Secondly, feeding larvae groups have successively greater mean shell lengths in relation to increasing particle size. Thus, the distributional data is suggestive of the first alternative hypothesis. Larvae fed a mixed suspension of Chlorella and Phaeodactylum consumed both algal species.

In this instance, larvae less than 120 μ in shell length took only the Chlorella. Larvae greater than 120 μ in shell length took both the Chlorella and the Phaeodactylum. However, larvae which fed on both species of algae apparently consumed the Phaeodactylum first: that is, larvae ingesting size dimorphic food had only Phaeodactylum in the digestive diverticulum while Chlorella was found only in the gut. Once again, it must be emphasized that the apparent differential treatment of food particles by A. papillosa veligers may be a function of mechanical limitation or selective size preference.

DISCUSSION

Development of both H. crassicornis and A. papillosa agree with the Type 1 (planktotrophic) development (Thompson, 1967). In each case, the ova are small and the egg capsules contain multiple embryos. Developmental time to hatching is relatively short. The free swimming veligers lack eyes, radula and propodial rudiment.

The early embryology and veliger morphology at hatching corresponds well with that of other cleioproct eolidoideans: Aeolidiella glauca Alder and Hancock, A. japonica Eliot, Facelina coronata (Forbes), and Favorinus branchialis (Müller) (Hadfield, 1963; Hamatani, 1967; Tardy, 1970; and Rasmussen, 1951, respectively). Of these species, only Aeolidiella japonica was observed to have sculpture patterns in the form of minute dots at the base of the aperture. However, unlike H. crassicornis and A. papillosa, veliger larvae belonging to the species cited above (exception: Aeolidiella japonica) demonstrated growth during the free-swimming stage.

Planktotrophic development and veliger morphology has been recently considered in three species of pleuroproct eolidoideans: Coryphella trilineata O'Donoghue (Bridges and Blake, In Press); and C. fusca O'Donoghue, and C. rufibranchialis (Johnston) (Hurst, 1967). Bridges and Blake observed holoblastic, unequal first cleavage for C. trilineata embryos, the formation of one to three polar bodies and rapid

development to hatching (5 days). Oblique striations were observed by Bridges and Blake and by Hurst on the ventral whorl of the coryphellid veliger shells. Shell growth, however, was not recorded.

Planktotrophic larvae among the acleioproct eolidoideans have been observed by Hurst (1967) for Eubranchus olivaceus (O'Donoghue), Catriona aurantia (Alder and Hancock) and Trinchesia albocrusta (MacFarland); and by Hadfield (1963) for Eubranchus exiguus. The striking difference between the acleioprocts as opposed to the pleuroprocts and cleioprocts is the development of a Type 2 (inflated) larval shell.

Examples of Type 2 (lecithotrophic) development among the Eolidoidea are the cleioproct, Aeolidiella mannarensis (Rao and Alagarswami), and the acleioproct, Cuthona adyarensis Rao (Rao and Alagarswami, 1960; Rao, 1961 respectively). Both species fulfill Thompson's (1967) criteria of lecithotrophic larvae but possess opposing Type 1 and Type 2 larval shells, respectively.

Excellent observations on direct development in the Eolidoidea are available in the work of Morse (1971) who described the life history of the pleuroproct, Coryphella stimpsoni (Verrill) and Tardy (1970) who examined the developmental and metamorphic features of the cleioproct, Aeolidiella alderi (Cocks).

Further attention will be paid to the developmental types within the Eolidoidea in subsequent portions of the discussion.

At present, it is noteworthy to point out that the genus Aeolidiella contains species with all three developmental types and that the genus Coryphella contains species with planktotrophic and direct development.

Functional Morphology and Feeding:

Thompson's (1959) comparative treatment of feeding behavior in nudibranch veligers concludes (p. 247) that, "the mechanism of feeding was very similar in all the species investigated. . ." The observations concerning ciliary mechanism and treatment of food in H. crassicornis and A. papillosa veligers are, with one exception, in agreement with Thompson. This exception is the transport of large undigested particles along the intestinal groove to the confluence of the dorsal gut and the intestine. This observation, however, agrees with Fretter and Montgomery's (1968) study of food treatment in 19 species of monotocardian prosobranchs.

Observations of ciliary metachronism were not included in Thompson's (1959) investigation of feeding. Metachronism in H. crassicornis and A. papillosa veligers concur with Fretter's (1967) detailed account of ciliation in prosobranch veligers.

The major differences between prosobranch veligers and nudibranch veligers are found in the anatomy of the gut, and in the passage of food into the diverticula. The prosobranch veliger has a folded gastric shield in the posteroventral gut wall (Fretter and Montgomery, 1968), while nudibranch veligers

have denticulate hyaline rods in the posteroventral gut wall (Thompson, 1959; present study). Thompson (1959) attributes a probable enzymatic function to the hyaline rods, similar to that of the style. Nudibranch veligers utilize only the left diverticulum in digesting food particles and the passage of food into the left diverticulum is achieved by ciliary means (Thompson, 1959; present study). Fretter and Montgomery, however, describe a muscular pumping action affecting ingestion and clearance of particles in both of the prosobranch diverticula.

Feeding Selectivity:

Before considering the specific case of H. crassicornis and A. papillosa feeding selectivity, some preliminary remarks concerning suspension feeding in general seem appropriate. In the definitive portions of Jorgensen's (1966) monograph on suspension feeding, emphasis is given to the distinctions between filter feeding, non-filter feeding, selective feeding and non-selective feeding; all under the auspices of suspension feeding. Thus, filter feeding is defined (p. xii) as "feeding by passing water through structures that retain particles mainly according to size and shape." Non-filter feeders (p. 131) are,

> "Those in which the water with its content of suspended particles is not truly filtered but is carried along surfaces capable of retaining particles that obtain contact with the surfaces. . . with emphasis placed on ciliary activity in renewing the water current along the particle collecting surfaces, as well as in carrying the collected particles to the mouth."

The distinction between selective and non-selective feeding falls in the restrictive use of the term "selection." Thus, the term selection applies to the qualitative sorting of food particles with respect to food value. In this sense, most suspension feeders are non-selective. On the other hand, selection in the broader sense of the term applies to the sorting of particles in relation to their size, shape or density. Thus, Jorgensen indicates that selection in the latter sense is better termed sorting mechanisms and is generally the rule as applied to suspension feeders. For purposes of this paper the term "selection" will be utilized in the broader sense with care given to making distinctions between particle quality or particle size, shape or density.

Size selectivity in filter feeding copepods is discussed by Gauld (1964) for Centropages and Calanus. In this instance selectivity is accomplished by differential utilization of the maxillae. Selectivity in non-filter feeding meroplanktonic forms is another matter. Qualitative selectivity is reported in veliger larvae of the bivalve, Mercenaria mercenaria (Loosanoff, Davis and Chanley 1953a, b; Davis and Loosanoff, 1953; Loosanoff, 1954). It was demonstrated under laboratory conditions that Porphyridium was of poor food value in culturing M. mercenaria larvae to metamorphosis. Secondly, when Porphyridium (3 μ) was offered in mixed suspension with Chlamydomonas (10 μ), the Porphyridium was rejected in favor of the Chlamydomonas. These observations, however, do not

obviate possible preferential treatment due to particle size.

Strathman (1971) offers an extensive coverage of suspension feeding behavior in 19 species of echinoderm larvae. Of particular interest are his observations on largest particles eaten, and interference with feeding by larger algae. Strathman shows that the largest particle length ingested by Ophiopholis and Strongylocentrotus droebachiensis 8 arm plutei is about 150 μ while that for Pisaster brachiolaria is about 210 μ. Also, for a given particle size, Ditylum (= 200 μ x 15-30 μ) larvae capable of ingesting smaller particles would eat only those Ditylum which were less than the mean length.

In the analysis of particle interference, Strathman fed Ophiopholis, Strongylocentrotus droebachiensis and Pisaster larvae single suspensions of Amphidinium and Ditylum, as well as, a mixed suspension of both Amphidinium and Ditylum. The number of Amphidinium ingested by larvae in mixed suspension was either unaffected (Pisaster) or reduced in comparison with the single suspensions. The number of the larger Ditylum ingested by larvae in mixed suspension was either unaffected (Ophiopholis and S. droebachiensis) or increased in comparison with the single suspension.

S. droebachiensis ate more Amphidinium than Ditylum in both single and mixed suspension. Ophiopholis ate more Amphidinium in single suspension, but more Ditylum in mixed. Pisaster ate equal amounts in single suspension but more Ditylum

in mixed suspension. Strathman concluded (p. 149) that, "Ditylum is cleared more efficiently but ejected from the buccal cavity, or alternatively, Amphidinium may be more easily ejected from the buccal cavity and, therefore, more Ditylum was eventually swallowed."

Secondly, "Pisaster apparently clears Ditylum more effectively but is stimulated to feed more by Amphidinium." These conclusions appear somewhat prejediced in favor of his hypothesis (p. 140) that, "if the feeding rate is lower in a given algal species on a mixture than it is on the species separately, then it is probably being rejected along with the less desirable or edible species in the mixture." That Ditylum is less desirable or less edible is an unsubstantiated value judgment predicated on its greater size. Granted that Amphidinium is being rejected, another conclusion may be that the larvae are disposed to efficiently consuming the largest particles possible. Thus, Ophiopholis and S. droebachiensis consume the smaller Ditylum, as well as, Amphidinium consequently reducing the feeding effort expressed by feeding on Amphidinium alone.

Selective feeding by veliger larvae is evaluated by Paulson and Scheltema (1968) for the prosobranch Nassarius obsoletus (Say). Their experiments were conducted in mixed suspensions of Phaeodactylum tricornutum, Cyclotella nana, and Dunaliella tertiolecta. Three size ranges of larvae were used; recently hatched, 200-400 μ, and 400-600 μ. They

conclude (pp. 485-488):

> "1.) Nassarius obsoletus veligers are able to feed selectively, 2.) there is an increasing preference for Phaeodactylum with increasing size, and 3.) the experiments do not reveal how the larvae select their food, whether it be by size, concentration, or chemotactile sense."

The apparent feeding behavior demonstrated by H. crassicornis and A. papillosa suggests that size of the food particle in relation to the veliger is a significant factor in selectivity. This, however, does not preclude possible effects due to algal concentration or food value (Loosanoff, Davis and Chanley, 1953a; Loosanoff, 1954; Calabrese and Davis, 1970; Pilkington and Fretter, 1970). Given identical concentrations of two or more different sized algal species, some obvious questions which come to mind are: what is the relative effect of volume displaced by each species on particle uptake? Secondly what are the relationships between clearance rate, ingestion rate and volumetric uptake for monomorphic, dimorphic and polymorphic algal suspensions? It is proposed, that the tentative conclusions pertaining to particle size selectivity be tested against varying known concentrations of algal suspensions. The problem of preferential or chemotactile selectivity can best be approached only when the possible effects of mechanical or particle size selectivity are evaluated.

Dimorphism and Larval Strategy:

The classification of H. crassicornis and A. papillosa as representatives of Thompson's (1969) planktotrophic development

has been established. It remains, however, to delineate the significance of the bimodal shell length distributions in relation to yolky and non-yolky larvae, arising from a single egg mass, a more speculative enterprise.

That the distributions are truly bimodal is accepted here on the basis of the graphic evidence. Corroborative evidence is the graphic representation of statistically different mean shell lengths of yolky and non-yolky veligers; each distributed normally. Secondly, the identity of yolky and non-feeding veligers is established for both species.

Thus, in the morphological context, A. papillosa and H. crassicornis veligers are planktotrophic larvae. However, if the presence of yolk reserves is not a teratological phenomenon, then yolky A. papillosa and H. crassicornis veligers may prove to be lecithotrophic in the metamorphic sense. In order to test these observations, however, it is necessary to establish the metamorphic substrate specificity for the veliger larvae in question. For the time being, however, the discussion will proceed on the assumption that the veliger larvae are dimorphic and that both yolky and non-yolky larvae are capable of progressive metamorphosis.

Thompson (1964, p. 275) has drawn the following generalizations from his observations on nudibranch life cycles:

> "Nudibranchs preying on organisms which form stable, abundant populations in certain types of locality have unusual life cycles and usually a single breeding period each year. Those which feed on more transitory prey usually pass through a number of generations each year and grow more

quickly to sexual maturity.

It is probably that new populations of nudibranchis are commonly established in favorable regions through the exercise of the ability of the veliger larvae to recognize and settle upon some component of the diet of the benthic stages."

Similarly, Miller (1962) notes that nudibranchs with several generations per year depend on fluctuating food supply which they exploit rapidly. He includes in this category all eolidoideans except the anemone eaters Aeolidia papillosa and Aeolidiella glauca.

Strathman (1971, p. 157) draws several interesting conclusions from his observations of echinoderm larvae.

"Echinoderms with planktotrophic larvae may, in each class, have evolved a manner of metamorphosis combining maximum preparation for a benthic existence with the ability to feed during the search for a suitable attachment site.

The asteroids which in general have the most restricted feeding habits, also undergo the least change in larval structures prior to settling."

Williams (1966, p. 70) states:

"A habitat is fit because its occupant provides itself with near-optimum soma for life in that particular habitat. The precision of this adaptation may be compromised by its future commitments. At any stage in its life history, an organism must not only adapt to its immediate circumstances, it must retain the ability to adapt to those likely to be encountered in the future."

With these generalities in mind and in the spirit of speculative enterprise we come to the case at hand. Both H. crassicornis and A. papillosa have spawn in the field throughout the year. Assuming, for the sake of further

argument, that phytoplankton populations fluctuate throughout the year, then the bimodal distributions of yolky and non-yolky larvae may be subjected to the following strategy.

1.) Yolked larvae will be successful in terms of survival in the plankton independent of food availability.

2.) Non-yolked larvae will be successful in terms of survival in a phytoplankton rich environment. Secondly, the food size selectivity in relation to larval size polymorphism offers a wide range of particle sizes which are acceptable to at least some of the non-yolked larvae. This is particularly applicable under conditions of phytoplankton succession.

Thus the selective pressures which produced all three developmental types in the genus Aeolidiella, and planktotrophic and direct developmental types within the genus Coryphella may very likely rest on the relationship between larval feeding ability and phytoplankton abundance.

In this respect it seems applicable to restate Hadfield's (1965, p. 94) conclusion that "probably most nudibranch veligers are quite plastic in terms of dependance on the planktonic period and length of time in the plankton, and are able to metamorphose or continue to swim from soon after hatching to an extended period."

LITERATURE CITED

Conover, Robert S. 1956. Oceanography of Long Island Sound, 1952-1954. VI. Biology of Aerstia claus and A. tonse. Bing. Oc. Coll. IV:156-234.

Calabrese, A. and H.C. Davis. 1970. Tolerances and requirements of embryos and larvae of bivalve molluscs. Helgolander Wiss. Meeresunters, Bd. 20(1-4):553-564.

Davis, Harry C. and Victor L. Loosanoff. 1953. Utilization of different food organisms by clam larvae. Anat. Rec. 117(3):646.

Fretter, V. 1967. The prosobranch veliger. Malacol. Soc. London, Proc. 37(6):357-366.

Fretter, Vera and Margaret C. Montgomery. 1968. The treatment of food by prosobranch veligers. J. Mar. Biol. Assoc. U.K. 48(2):499-520.

Gauld, D.T. 1964. Feeding in planktonic copepods. Brit. Ecol. Soc. Symp. 4: Grazing in Terrestrial and Marine Environments. D.J. Crisp, Ed. Blackwell. Oxford. pp. 239-245.

Guillard, R.R.L. and J.H. Ryther. 1962. Studies on marine plankton diatoms I. Cyclotella nana Hustedt and Detonula conferuacea (Cleve). Can. J. Microbiol., Trans. 8:229-239.

Hadfield, Michael G. 1963. The biology of nudibranch larvae. Oikos. 14(1):85-95.

Hamatani, Iwao. 1967. Notes on veligers of Japanese opisthobranchs (7). Publ. Seto. Mar. Biol. Lab. 15(2):121-131.

Hurst, Ann. 1967. The egg masses of veligers of thirty Northeast Pacific opisthobranchs. Veliger 9(3):255-287.

Jorgensen, C. Barker. 1966. Biology of Suspension Feeding. Int. Ser. Mon Pure App. Biol. 27. Pergamon Press. London. 357 p.

Knight-Jones, E.W. 1954. Relations between metachronism and the direction of ciliary beat in metazoa. Quart. J. Microsc. Sci. 95(4):503-521..

Loosanoff, Victor L. 1954. New advances in the study of bivalve larvae. Am. Scien. 42(4):607-624.

Loosanoff, Victor L., H.C. Davis and Paul E. Chanley. 1953a. Behavior of clam larvae in different concentrations of food organisms. Anat. Rec. 117(3):586-587.

_____ 1953b. Effect of overcrowding on rate of growth of clam larvae. ibid. pp. 645-646.

Marshall, R.M. and A.P. Orr. 1964. Grazing by copepods in the sea. Brit. Ecol. Soc. Symp. 4: Grazing on Marine and Terrestrial Environments. D.J. Crisp, Ed. Blackwell. Oxford. pp. 227-237.

Miller, Michael Charles. 1962. Annual cycles of some Manx nudibranchs with a discussion of the problem of migration. J. Anim. Ecol. 31:545-569.

Morse, M. Patricia. 1971. Biology and life history of the nudibranch mollusc, Coryphella stimpsoni (Verrill, 1879).. Biol. Bull. 140(1):84-94.

Paulson, T.C. and R.S. Scheltema. 1968. Selective feeding on algal cells by the veliger larvae of Nassarius obsoletus (Gastropoda, Prosobranchia). Biol. Bull. Mar. Biol. Lab., Woods Hole. 134:481-489.

Pilkington, M.C. and V. Fretter. 1970. Some factors affecting the growth of porsobranch veligers. Helgolander Wiss. Meeresunters, Bd. 20(1-4):576-593.

Rao, K. Virabhadra. 1962. Development and life history of a nudibranchiate gastropod Cuthona adyarensis Rao. J. Mar. Biol. Ass. India 3(1-2):186-197.

Rao, Virabhadra K. and K. Algarswami. 1960. An account of the structure and early development of a new species of a nudibranchiate gastropod Eolidina (Eolidina) mannarensis. J. Mar. Biol. Ass. India 2(1):6-17.

Rasmussen, Erik. 1951. Faunistic and biological notes on marine invertebrates II. The eggs and larvae of some Danish marine gastropods. Vid. Midd. Dansk. Naturh. Foren. Bd. 113:201-249.

Raven, Chr. P. 1958. Morphogenesis: The Analysis of Molluscan Development. Int. Ser. Monog. Pure Appl. Biol. 2. Pergamon Press. London. 311 p.

Sokal, R.R. and F.J. Rohlf. 1969. Biometry, the Principles and Practice of Statistics in Biological Research, W. H. Freeman and Co. San Francisco. 776 p.

Strathman, Richard B. 1971. The feeding behavior of planktotrophic echinoderm larvae: Mechanisms, regulation and rates of suspension feeding. J. Exp. Mar. Biol. and Ecol. 6(2):109-160.

Tardy, Jean. 1970. Contribution a l'etude des metamorphoses chez les nudibranchis. Zoologie et biologie animale 12(3):299-371.

Thompson, T.E. 1958. The natural history, embryology, larval biology and post-larval development of Adalaria proxima (Alder and Hancock), (Gastropoda, Opisthobranchia). Roy. Soc. London, Phil. Trans. B 242:1-58.

_____ 1959. Feeding in nudibranch larvae. J. Mar. Biol. Ass. U.K. 38:239-248.

_____ 1961. The importance of the larval shell in the classification of the Sacoglossa and the Acoela. Malacol. Soc. London, Proc. 34(5):233-238.

_____ 1962. Studies on the ontogeny of Tritonia hombergi Cuvier (Gastropod, Opisthobranchia). Roy. Soc. London, Phil. Trans. B 245:171-218.

_____ 1967. Direct development in a nudibranch, Cadlina laevis with a discussion of developmental processes in Ophisthobranchia. J. Mar. Biol. Ass. U.K. 47:1-22.

Tinkle, Donald W. 1969. The concept of reproductive effort and its relation to the evolution of life histories of lizards. Am. Nat. 103(933):501-516.

Williams, George C. 1966. Adaptation and Natural Selection: A Critique of Some Current Evolutionary Thought. Princeton University Press. Princeton, New Jersey.

Williams, Leslie G. (In press). Veliger development in Dendronotus frondosus (Ascanius, 1774), (Gastropoda, Nudibranchia). The Veliger.

Table I

Developmental rate to hatching for *H. crassicornis* and *A. papillosa* cultured in seawater at 14.0°C.

Stage	Cumulative Hours	
	H. crassicornis	*A. papillosa*
1st polar body	1.5	2.0
2nd polar body	3.4	5.0
1st cleavage	4.9	6.0
2nd cleavage	6.1	7.5
3rd cleavage	7.0	9.5
4th cleavage	9.3	13.0
Morula	12.5	25.0
Blastula	23.7	31.0
Early gastrula	31.0	45.0
Late gastrula	51.0	51.0

Table I (Continued)

Trochophore	65.0	82.0
Shell gland invagination	75.0	88.0
Shell gland evagination	83.0	95.0
Early veliger	101.1	118.0
Alimentary differentiation	135.0	160.0
Hatching	166.0	195.0

Table II

Veliger shell measurements for H. crassicornis and A. papillosa

	H. crassicornis			A. papillosa		
	N	$\bar{X}$	S	N	$\bar{X}$	S
Length	200	103.82μ	8.46	200	116.18μ	10.11
Height	27	82.94μ	9.88	30	87.85μ	8.89
Width	39	80.65μ	6.05	30	85.90μ	7.66

Table III

Mean diameters (length for *Dunaliella*) and standard deviations of algal species accepted as food by veligers.

N = 100

Species	$\bar{X}$	S
Monochrysis lutheri	4.1 μ	± 1.2
Isochrysis galbana	4.2 μ	± 1.4
Chlorella 580	4.4 μ	± 1.1
Dunaliella tertiolecta	9.4 μ	± 1.6

Table IV

H. crassicornis

Analysis of variance. Significant added variance component among all groups with $p < 0.001$.

A priori comparisons among two means

with $F_{0.05(1,114)} = 3.93$.

N = 20

Source of variation	MS	F_s	p
Yolky vs. Non-yolky	907	22.84	p 0.001 ***
Feeding vs. Non-feeding	1,048	26.39	p 0.001 ***
Non-yolky vs. Feeding	73	1.84	0.25 p 0.10
Yolky vs. Non-feeding	1	0.03	1.00 p 0.75
Feeding Chlorella vs. Feeding Dunaliella	203	5.11	0.05 p 0.025 *

Table V

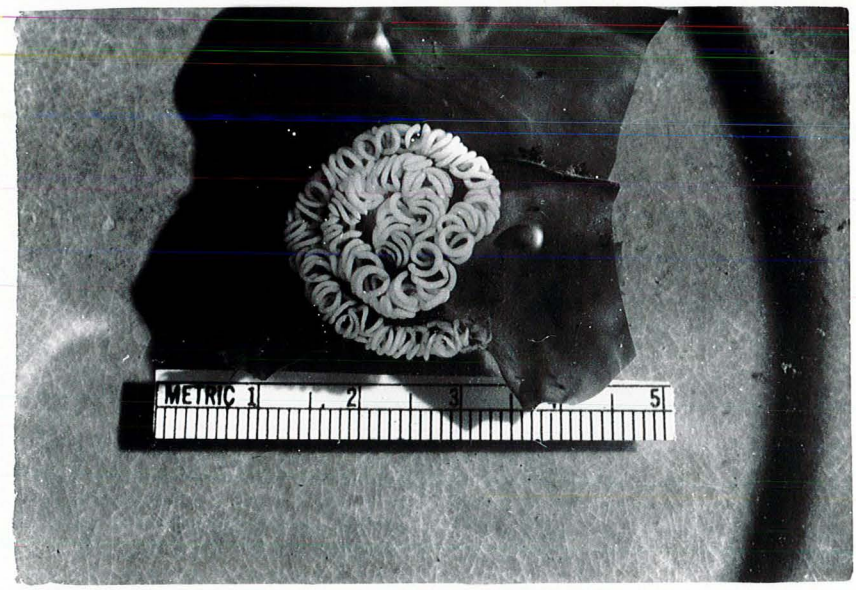
A. papillosa

Analysis of variance. A priori comparisons among two means with $F_{0.05(1,301)} = 3.84$

Source of variation	MS	F_s	p
Yolky vs. Non-yolky	4,409.0	64.28	p 0.001 ***
Non-yolky vs. Feeding	1,128.0	16.44	p 0.001 ***
Yolky vs. Non-feeding	18.5	0.26	0.500 p 0.750
Feeding vs. Non-feeding	8,344.0	121.66	p 0.001 ***
Feeding *Chlorella* vs. Feeding *Dunaliella*	205.0	2.99	0.050 p 0.100
Feeding *Dunaliella* vs. Feeding *Phaeodactylum*	135.0	1.96	0.100 p 0.250
Feeding *Chlorella* vs. Feeding *Phaeodactylum*	673.0	9.81	0.001 p 0.005**
Feeding Dimorphic vs. Feeding (*Chlorella* and *Phaeodactylum*)	22.1	0.32	0.500 p 0.750

Figure 1. H. crassicornis egg mass.

Figure 2. A. papillosa egg string with capsules joined by twisted portion of capsule membrane. Scale 100 μ.


METRIC 1
2
3
5

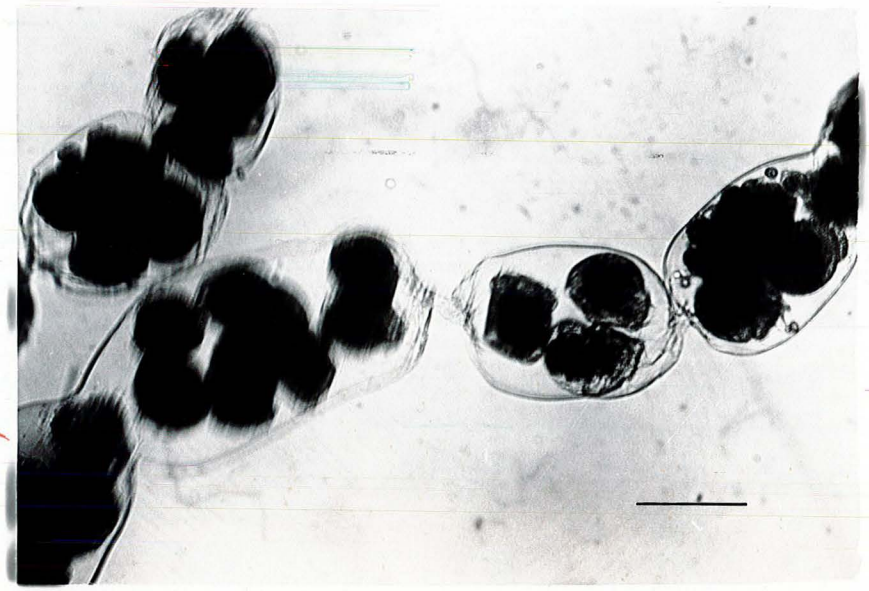


Figure 3. Means and 95% confidence intervals for H. crassicornis and A. papillosa zygote diameters. Coefficient of intraclass variation is 0.950.

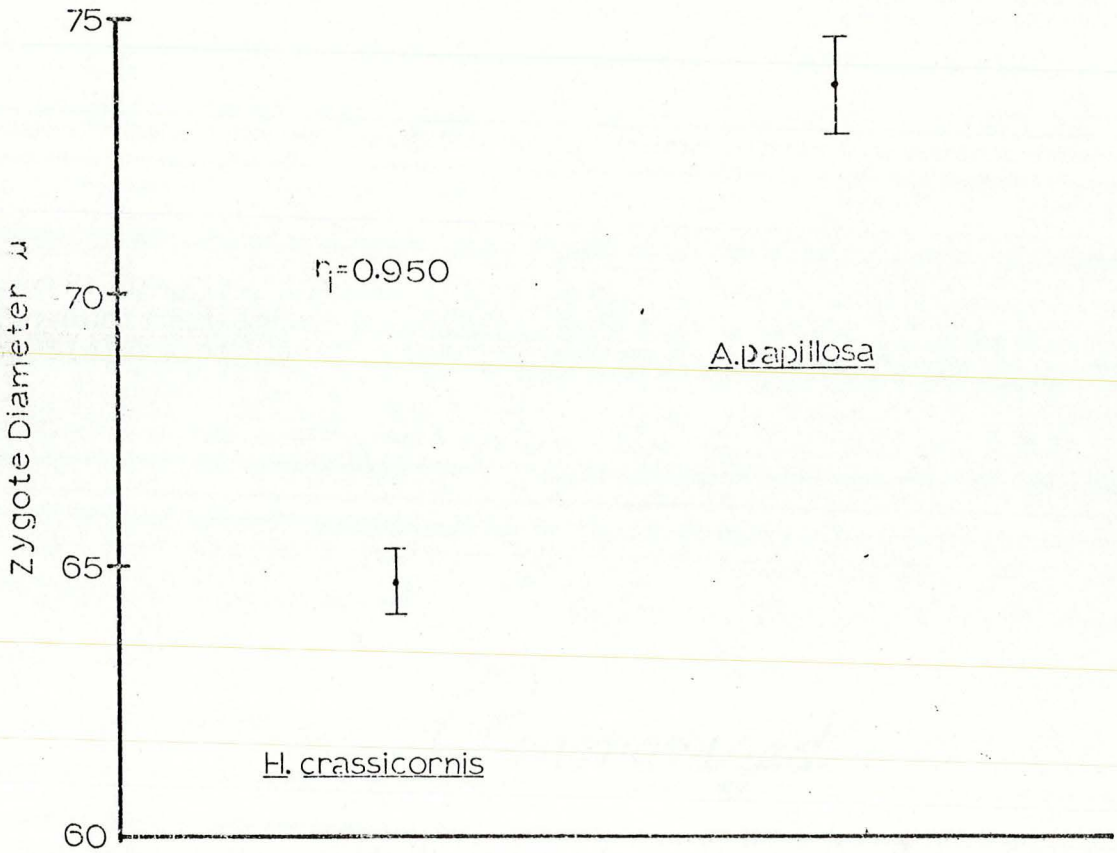
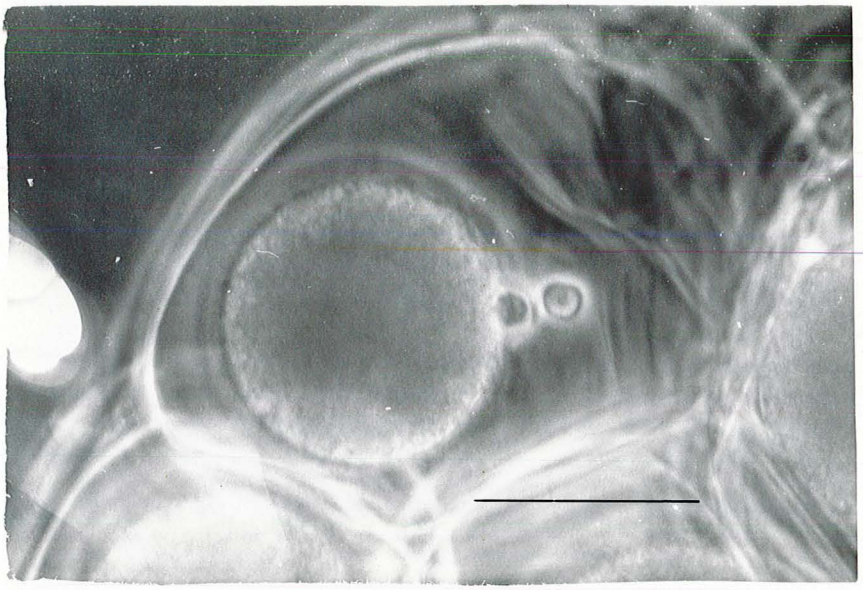

75
70
65
60
Zygote Diameter μ
η=0.950
A.papillosa
H. crassicornis

Figure 4a. H. crassicornis zygote and two polar bodies. Scale 50 μ.

Figure 4b. A. papillosa zygote and two polar bodies. Arrow indicates cleavage furrow of first polar body. Scale 50 μ.



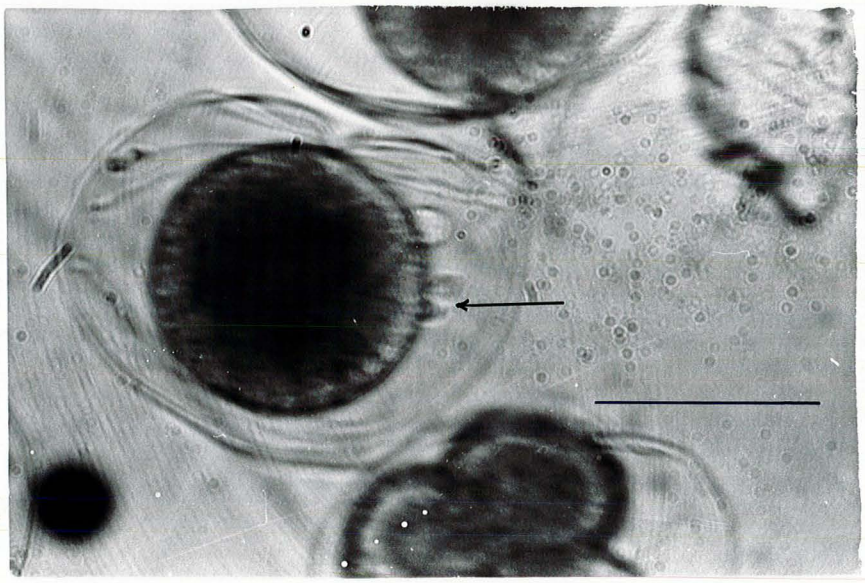
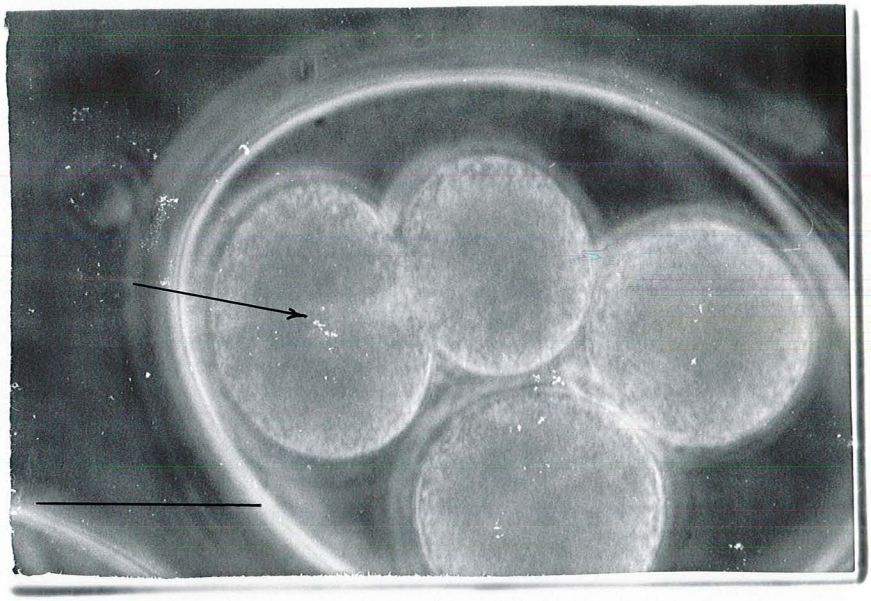


Figure 5. H. crassicornis at beginning of second cleavage. Arrow indicates cleavage furrow during formation of three cell stage. Scale 50 μ.

Figure 6. H. crassicornis stereoblastula with polar bodies at animal pole. Scale 50 μ.



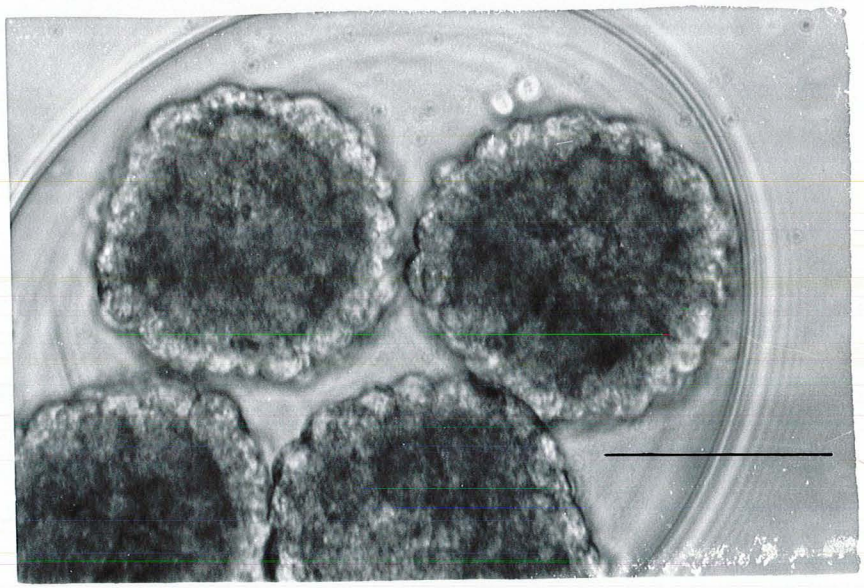
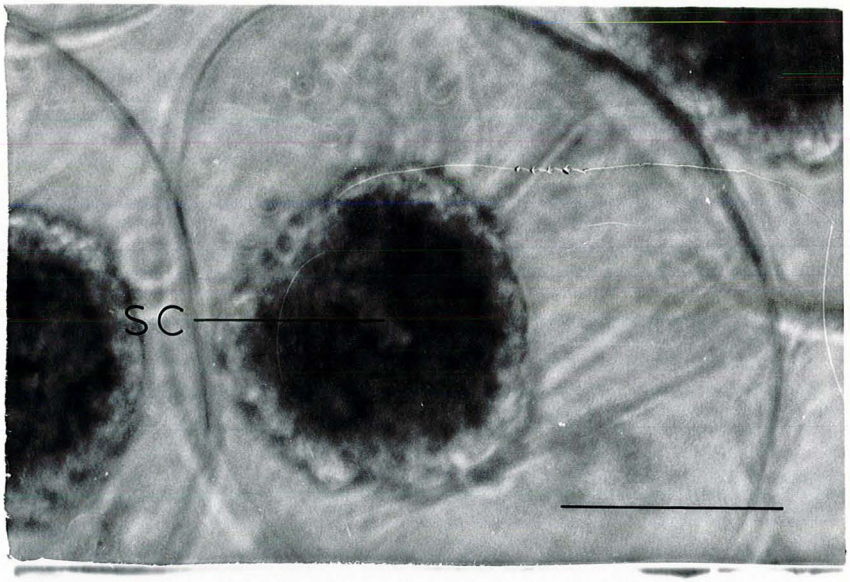


Figure 7. H. crassicornis stereogastrula; SC, sagittal cleft. Scale 50 μ.

Figure 8. H. crassicornis trochophore; SGI, shell gland invagination. Scale 50 μ.


SC


SGI

Figure 9. H. crassicornis free swimming veliger. ap. cl., apical cilia; int., intestine; ldd., left digestive diverticulum; lk., larval kidney; mf, mantle fold; mm., mantle muscle; op., operculum; rdd., right digestive diverticulum; st., statocyst; stom., stomach; str., striations; vel., velar cilia.

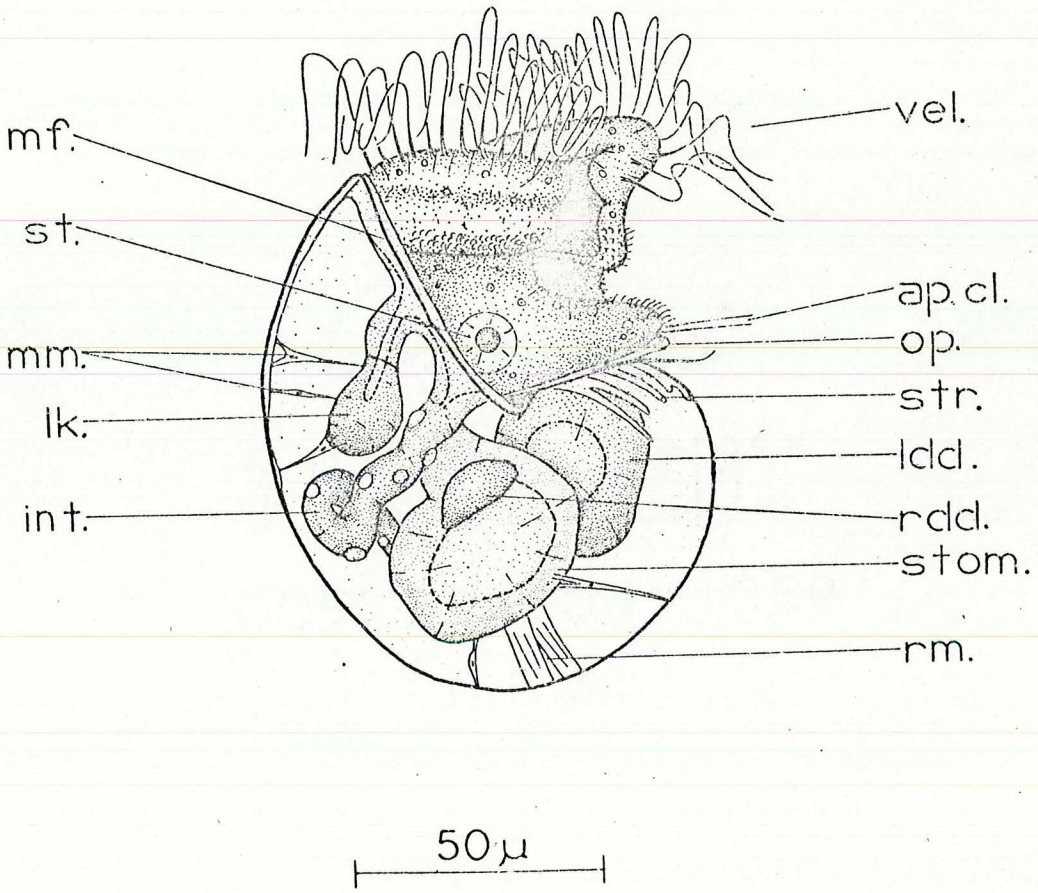

vel.
mf.
st.
ap. cl.
op.
mm.
str.
lk.
ldd.
int.
rdd.
stom.
rm.
50 μ

Figure 10. Larval shell length frequency distribution. Number of larvae plotted against shell length. Top, A. papillosa; bottom, H. crassicornis.

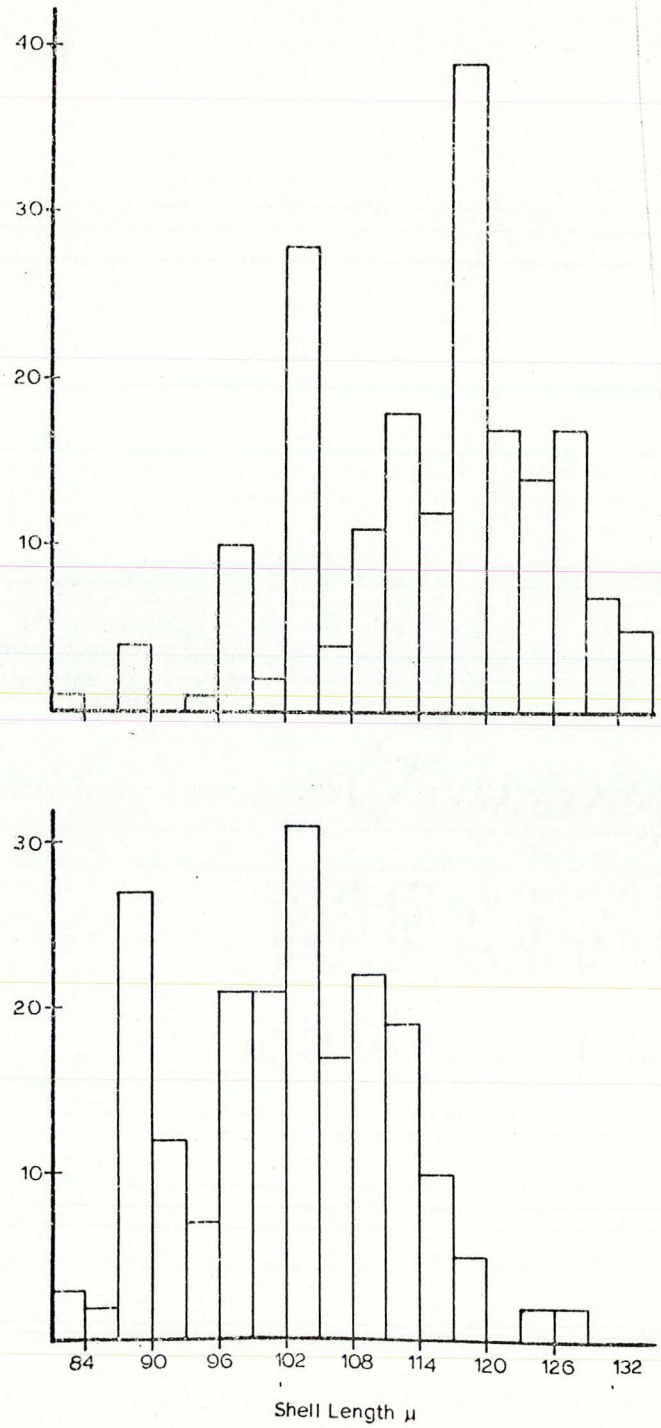

40
30
20
10
30
20
10
84
90
96
102
108
114
120
126
132
Shell Length μ

Figure 11. Larval shell length frequency distribution. Cumulative percent in probability scale plotted against shell length.

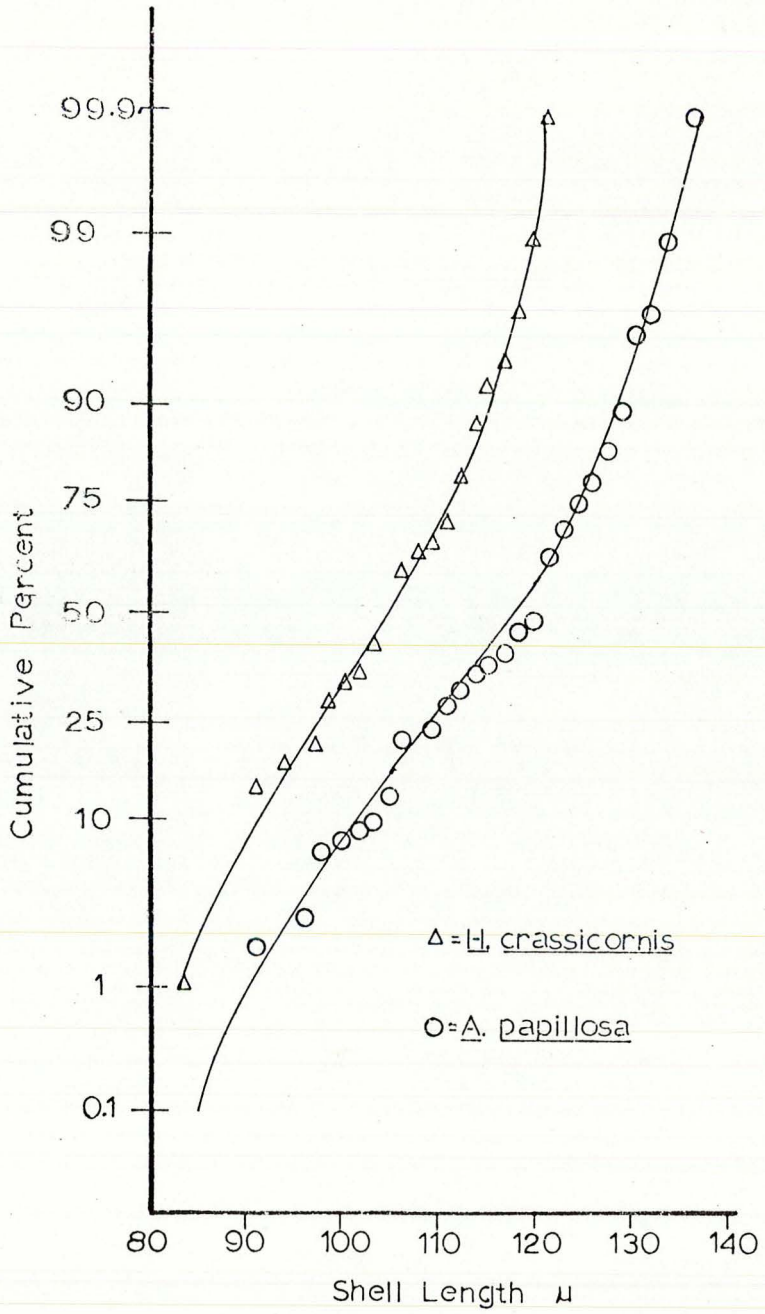

99.9
99
90
75
50
25
10
1
0.1
Cumulative Percent
80
90
100
110
120
130
140
Shell Length μ
Δ = H. crassicornis
O = A. papillosa

Figure 12. Schematic of H. crassicornis veliger. Solid arrow indicates direction of metachronal wave pattern in velar cilia. Dashed arrow shows direction of the effective stroke of post oral cilia.

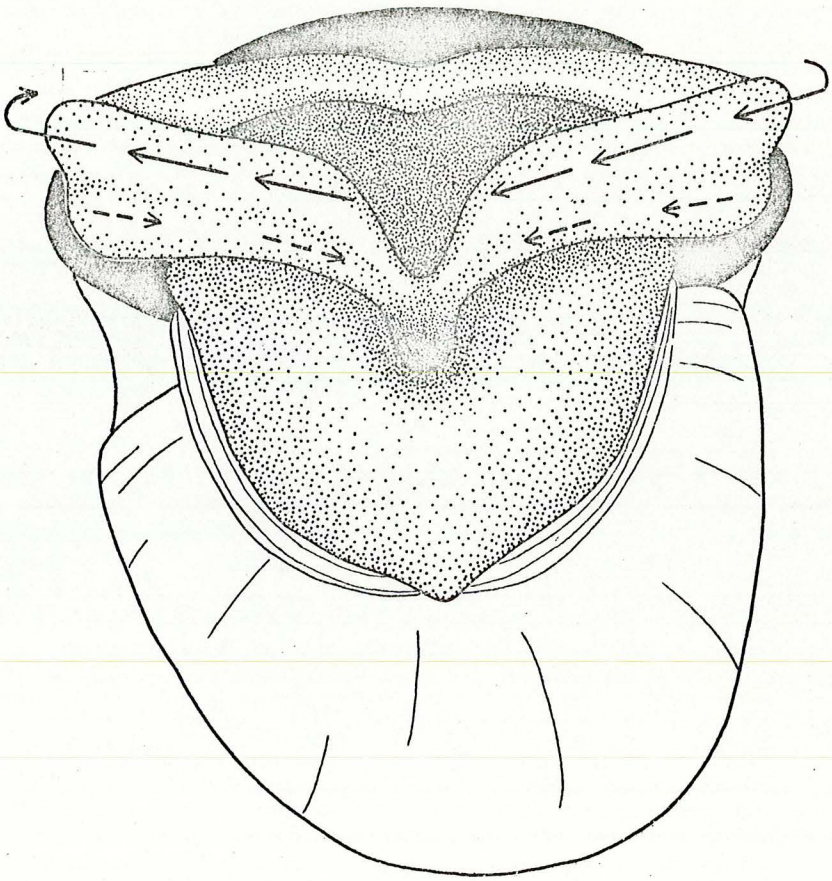


Figure 13. H. crassicornis veliger. Rotational movement. Arrow indicates contracted right velar lobe. Scale 50 μ.

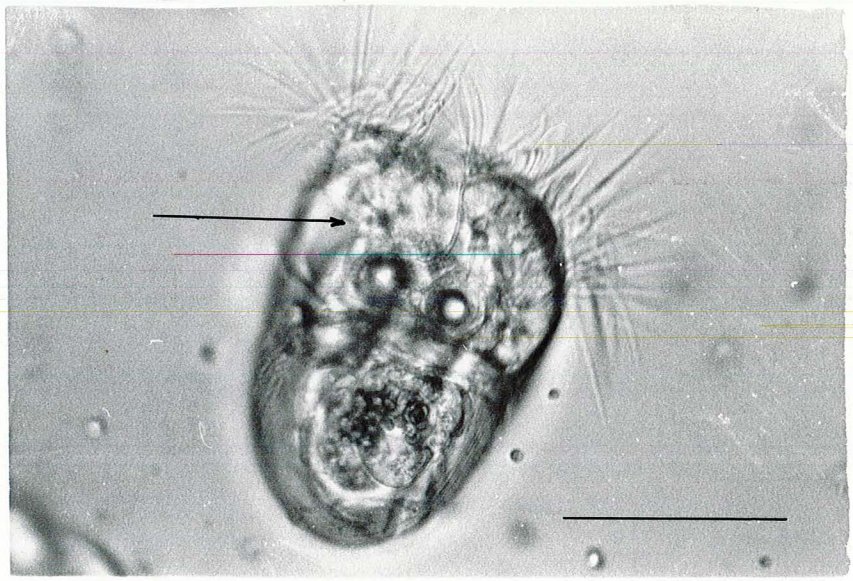


Figure 14. Schematic representation of veliger cephalopedal region showing ciliary currents affected by the velar, post oral and pedal cilia.

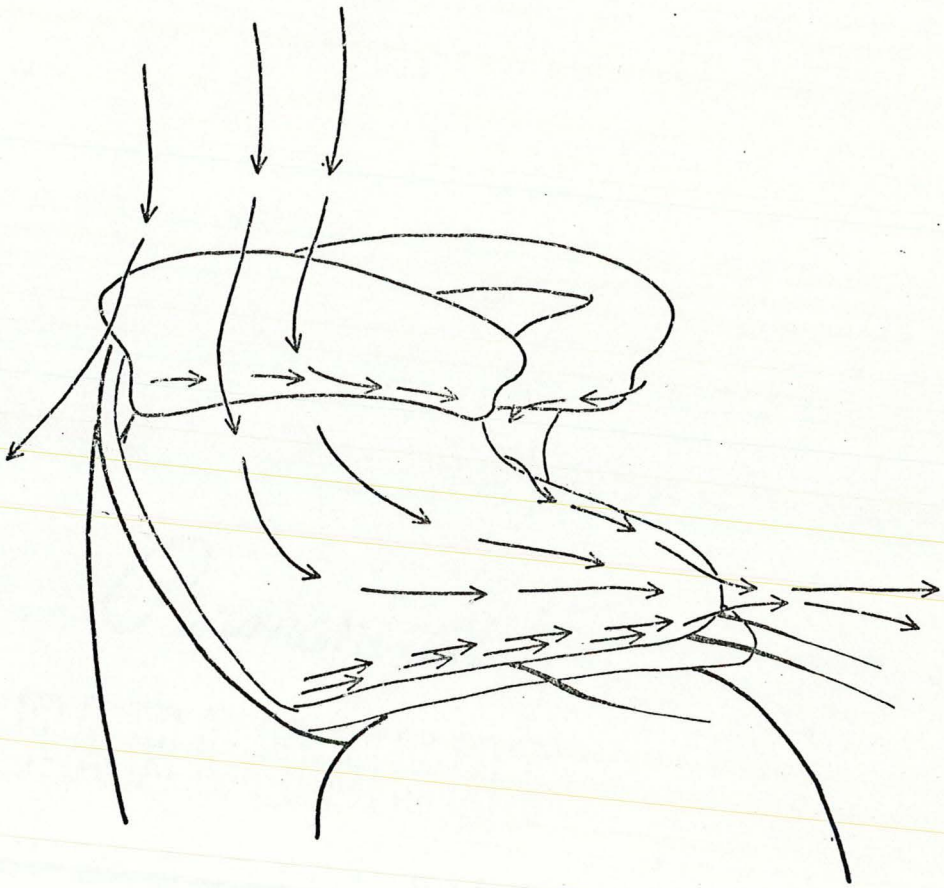


Figure 15. Schematic of the larval gut. Left figure shows direction of particle movement and denticulate hyaline rods. Right figure shows direction of particle movement via the intestinal groove.

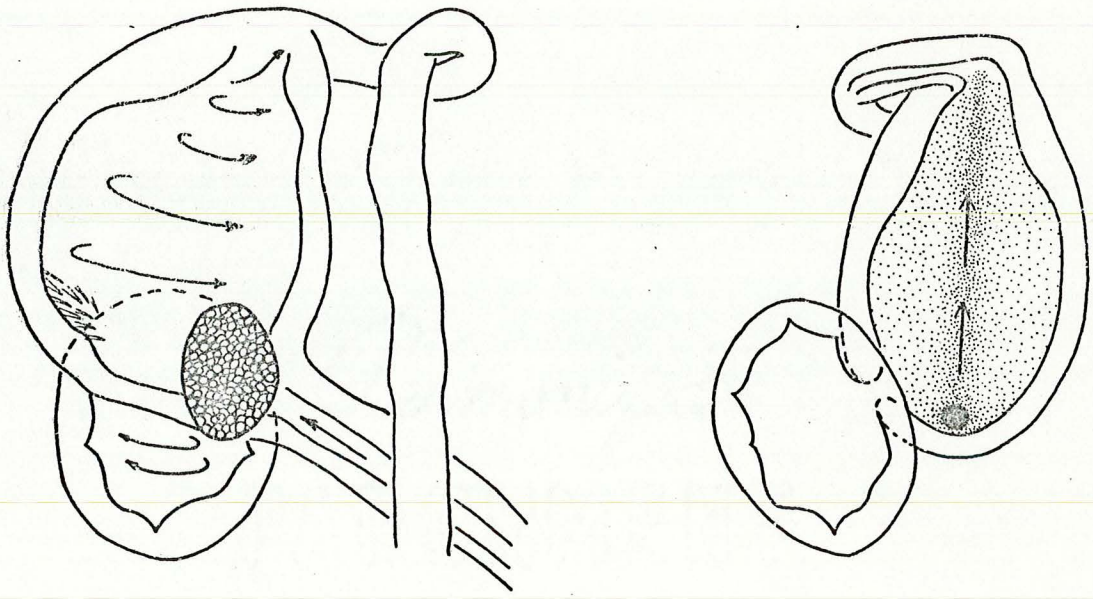


Figure 16. Percent number of larvae with algal particles in gut and/or diverticula plotted against number of hours after removal from a mixed algal suspension.

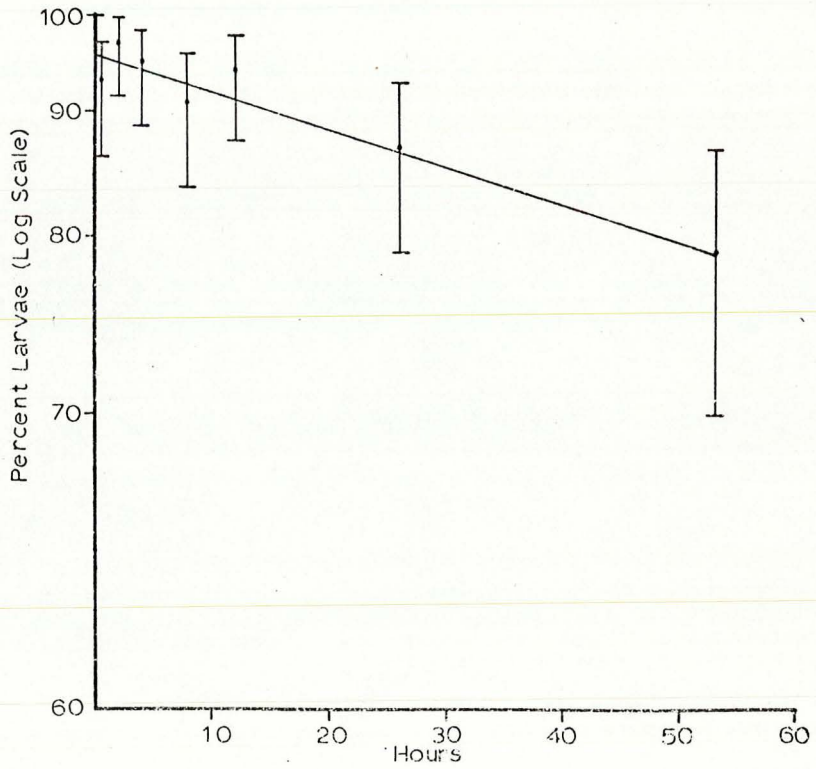

100
90
80
70
60
Percent Larvae (Log Scale)
10
20
30
40
50
60
Hours

Figure 17. Percent non-yolky and percent feeding larvae with 95% confidence intervals. fC, feeding on Chlorella; fC+P, feeding on both Chlorella and Phaeodactylum; fD, feeding on Dunaliella; fP, feeding on Phaeodactylum, ny, non-yolky larvae; (M), mixed suspension; (S), unialgal suspension.

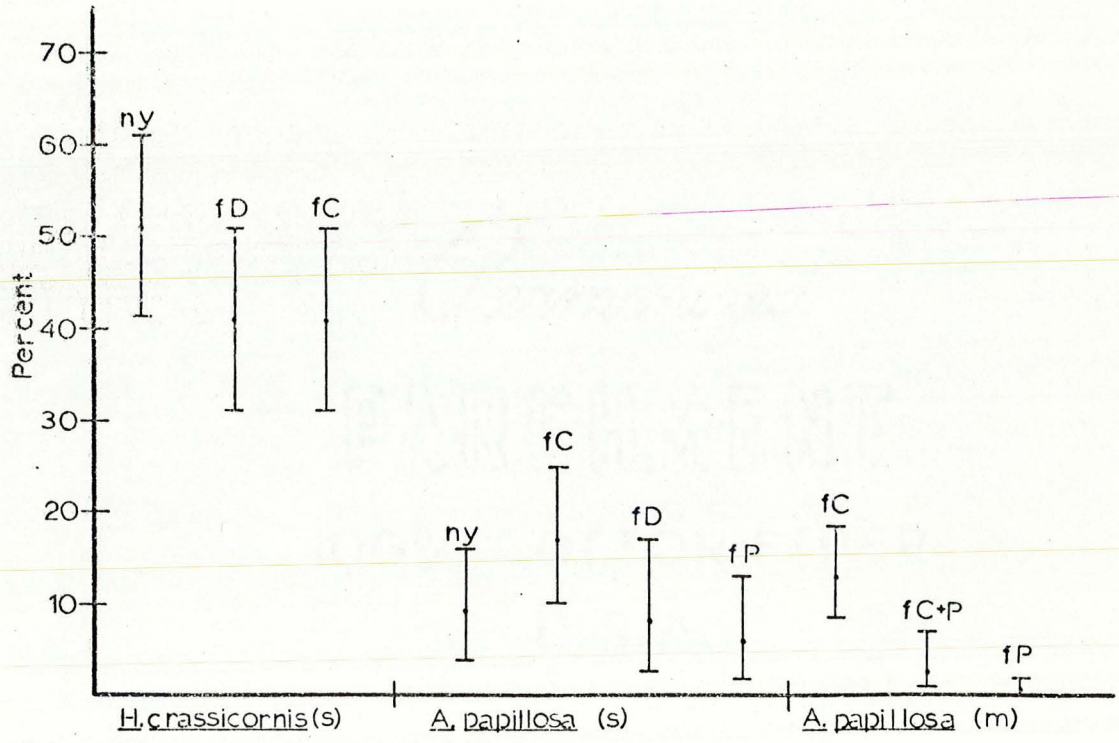

70
60
50
40
30
20
10
Percent
ny
fD
fC
ny
fC
fD
fP
fC
fC+P
fP
H. crassicornis (s)
A. papillosa (s)
A. papillosa (m)

Figure 18a. H. crassicornis . Means and 95% confidence intervals for shell length of experimental veligers. fC, larvae feeding in Chlorella suspension; fD, larvae feeding in Dunaliella suspension; nfC, larvae not feeding in Chlorella suspension; nfD, larvae not feeding in Dunaliella suspension; nY, non-yolky larvae; RS, random sample (N=100); Y, yolky larvae.

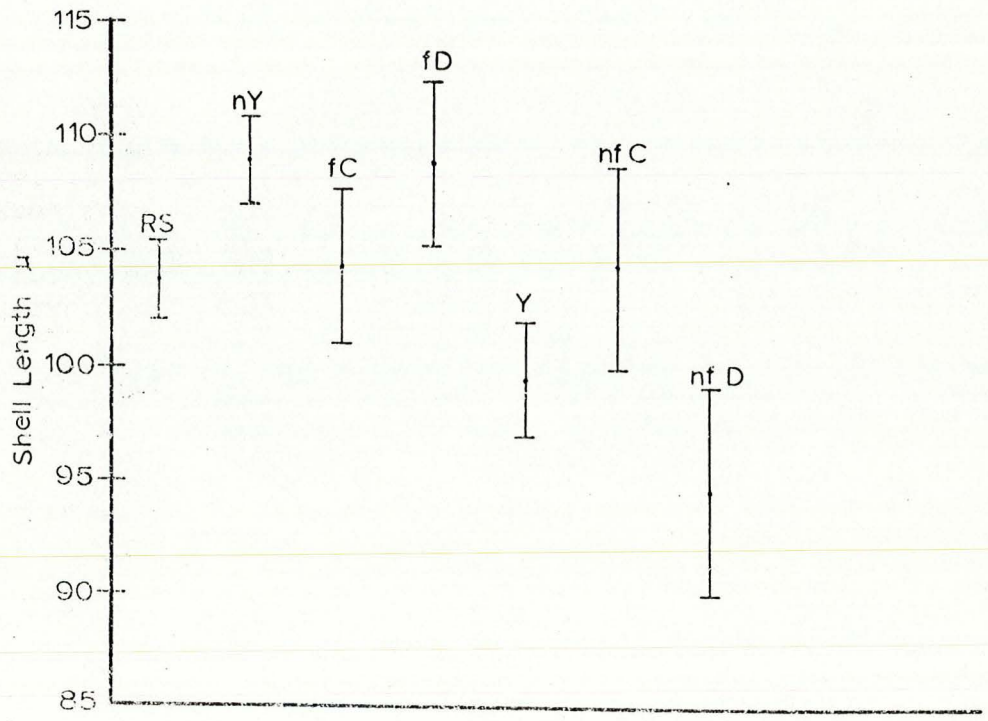

115
110
105
100
95
90
85
Shell Length μ
RS
nY
fC
fD
Y
nf C
nf D

Figure 18b. *A. papillosa*. Means and 95% confidence intervals for experimental veligers. fC, larvae feeding in *Chlorella* suspension; fD, larvae feeding in *Dunaliella* suspension; fP, larvae feeding in *Phaeodactylum* suspension; f(C+P), larvae feeding in combined suspension of *Chlorella* and *Phaeodactylum*; nf C, larvae not feeding in *Chlorella* suspension; nf D, larvae not feeding in *Dunaliella* suspension; nf P, larvae not feeding in *Phaeodactylum* suspension; nY, non-yolky larvae; RS, random sample (N=100); Y, yolky larvae.

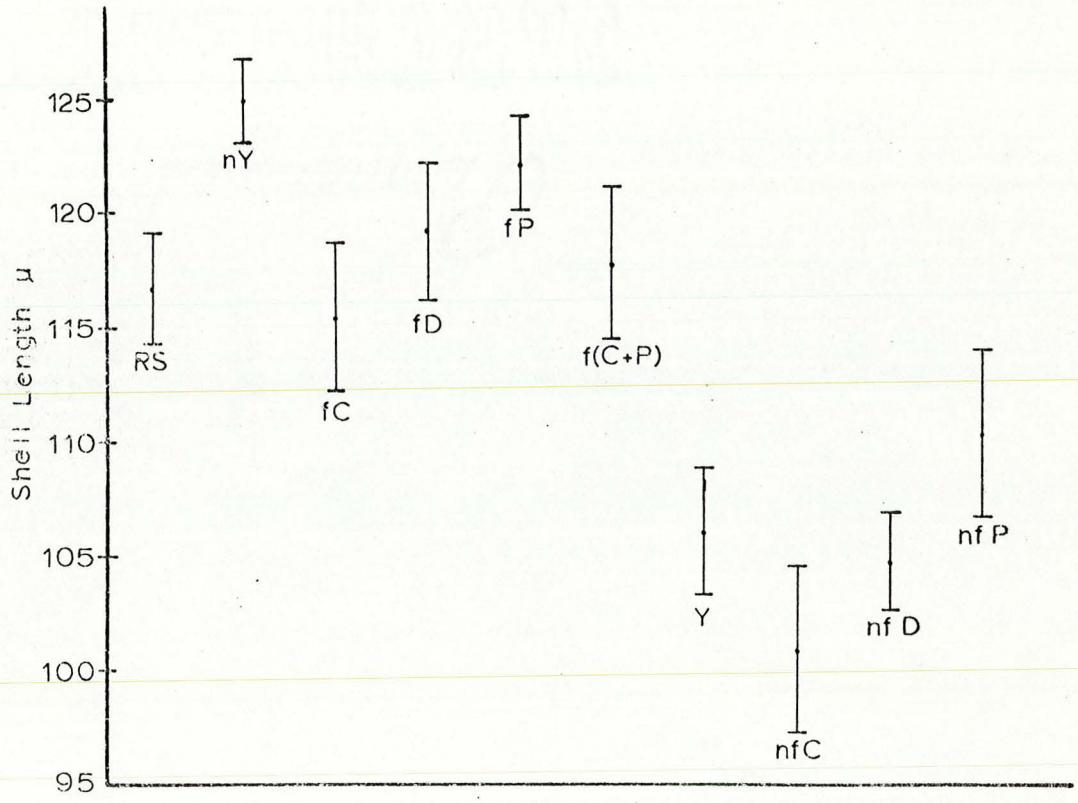

Shell Length μ
125
120
115
110
105
100
95
RS
nY
fC
fD
fP
f(C+P)
Y
nfC
nfD
nfP

Figure 19. Shell lengths of yolky and non-yolky larvae plotted against Rankits. Circles, yolky larvae; triangles, non-yolky larvae. Solid symbols and dashed lines, H. crassicornis. Open symbols and solid lines, A. papillosa.

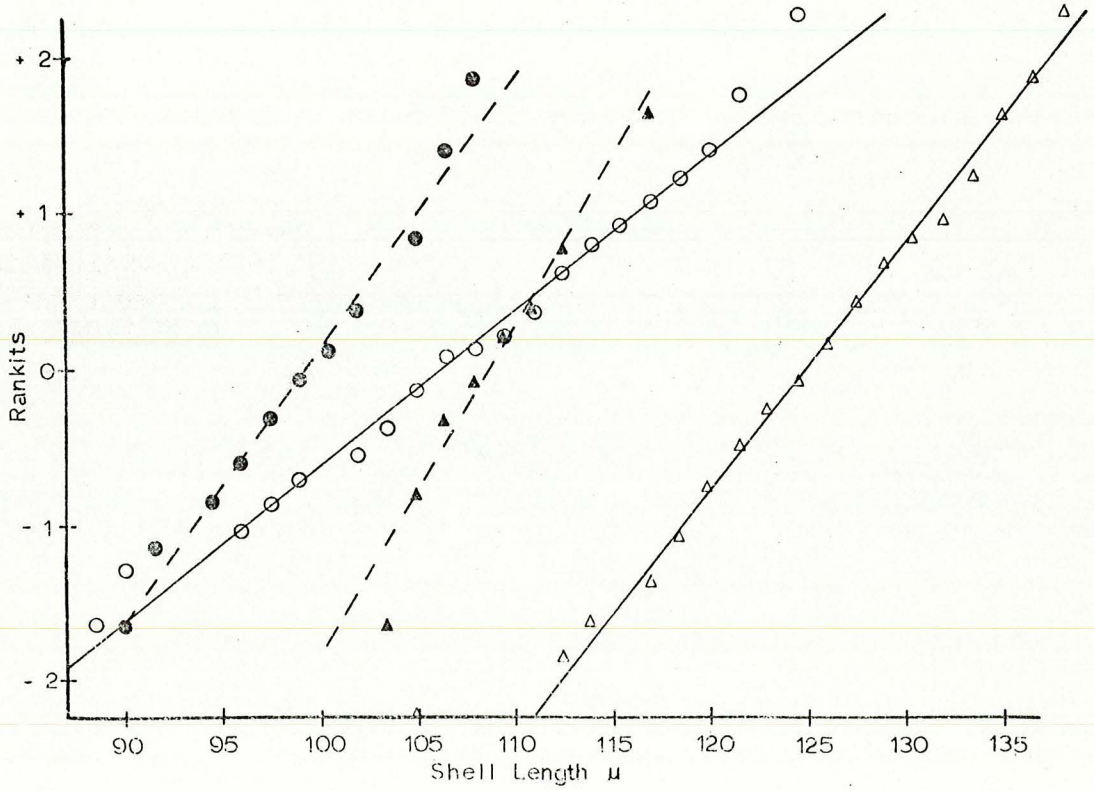

+ 2
+ 1
0
- 1
- 2
Rankits
90
95
100
105
110
115
120
125
130
135
Shell Length μ

Figure 20. H. crassicornis. Shell lengths of feeding and non-feeding larvae plotted against Rankits. Solid symbols and dashed lines represent non-feeding larvae. Open symbols and solid lines represent feeding larvae. Circles, Dunaliella suspension; triangles, Chlorella suspension.

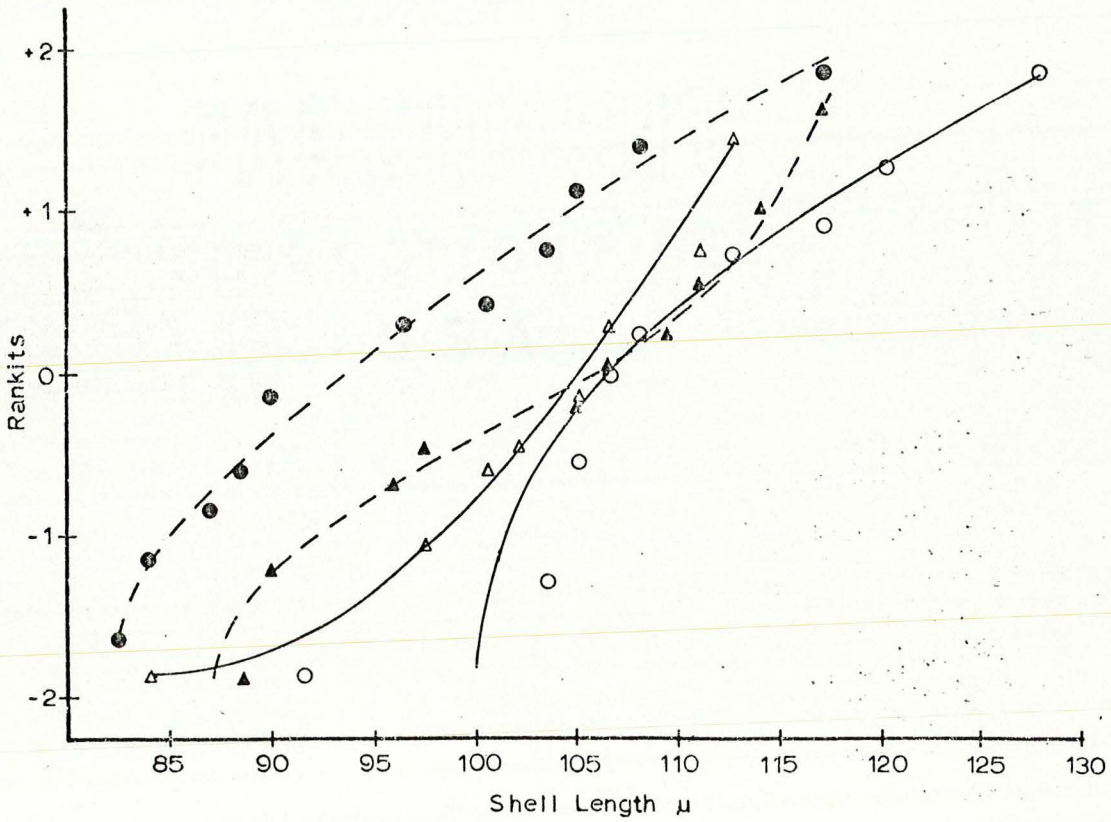
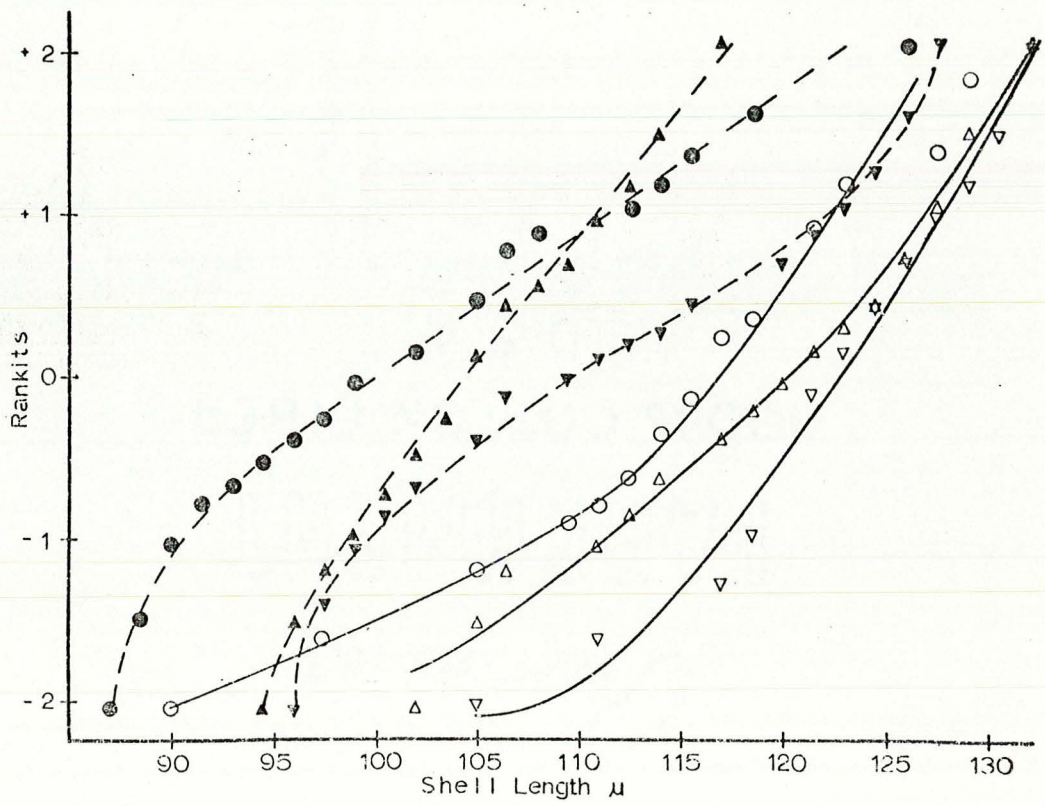

+2
+1
0
-1
-2
Rankits
85
90
95
100
105
110
115
120
125
130
Shell Length μ

Figure 21. A. papillosa. Shell lengths of feeding and non-feeding larvae plotted against Rankits. Solid symbols and dashed lines represent non-feeding larvae. Open symbols and solid lines represent feeding larvae. Circles, Chlorella suspension; triangles, Dunaliella suspension; inverted triangles, Phaeodactylum suspension.


+ 2
+ 1
0
- 1
- 2
Rankits
90
95
100
105
110
115
120
125
130
Shell Length μ